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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 5 : C07H 19/01, A61K 31/70 — A1

(11) International Publication Number: WO 91/13899

(43) International Publication Date: 19 September 1991 (19.09.91)

(21) International Application Number: PCT/JP91/00314

(22) International Filing Date: 8 March 1991 (08.03.91)

(30) Priority data:
9005521.1 12 March 1990 (12.03.90) GB
9017450.9 9 August 1990 (09.08.90) GB

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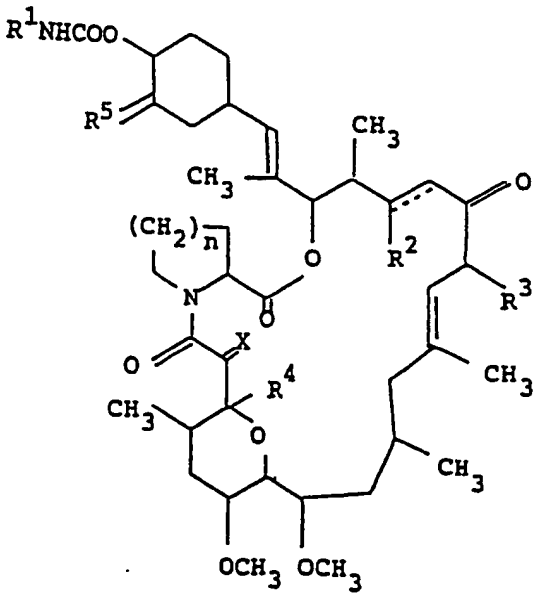
(81) Designated States: AT (European patent), BE (European patent), CH (European patent), DE (European patent), DK (European patent), ES (European patent), FR (European patent), GB (European patent), GR (European patent), IT (European patent), JP, LU (European patent), NL (European patent), SE (European patent), US.

Published

With international search report.

Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: TRICYCLO COMPOUNDS



(I)

(57) Abstract

Compounds of formula (I) are disclosed, wherein R^1 is hydrogen, lower alkyl which may have one or more hydroxy, or aryl which may have suitable substituent(s), R^2 is hydrogen, hydroxy or protected hydroxy, R^3 is methyl, ethyl, propyl or allyl, R^4 is hydroxy or alkoxy, R^5 is oxo, (H, OH) or (H, alkoxy), X is oxo or (H, OH), n is an integer of 1 or 2, and the symbol of a line and dotted line is a single bond or a double bond, and salts thereof. And processes for their production, compositions containing them, and use as immunosuppressive agents are also described.

DESCRIPTION

TRICYCLO COMPOUNDS

This invention relates to novel tricyclo compounds having pharmacological activities, to a process for their production and to a pharmaceutical composition containing the same.

More particularly, it relates to novel tricyclo compounds, which have pharmacological activities such as immunosuppressive activity, antimicrobial activity, and the like, to a process for their production, to a pharmaceutical composition containing the same and to a use thereof as a medicament.

Accordingly, one object of this invention is to provide the novel tricyclo compounds, which are useful as an immunosuppressant and an antimicrobial agent.

Another object of this invention is to provide a process for production of the tricyclo compounds by synthetic process.

A further object of this invention is to provide a pharmaceutical composition containing, as active ingredients, the tricyclo compounds.

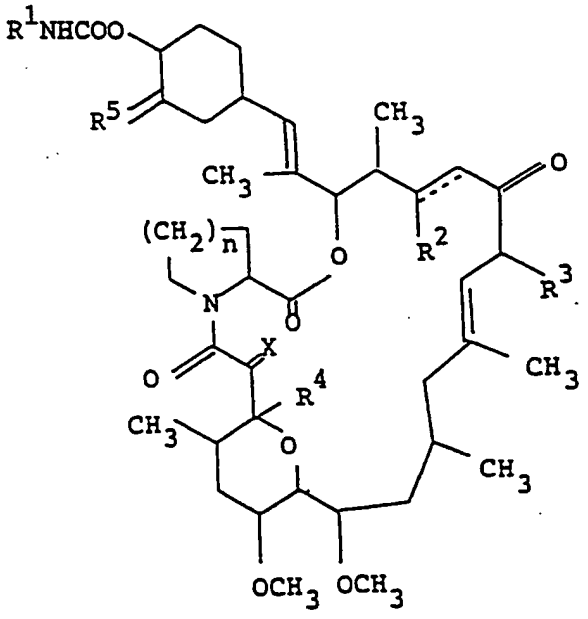
Still further object of this invention is to provide a use of the tricyclo compounds as a medicament for treating and preventing immune-mediated diseases such as resistance by transplantation, graft-versus-host diseases by medulla ossium transplantation, autoimmune diseases and the like, and further infectious diseases.

European Patent Application 184162 (Fujisawa Pharmaceutical Co. Ltd.) discloses a number of macrocyclic compounds isolated from microorganisms belonging to genus Streptomyces such as Streptomyces tsukubaensis No. 9993 (FERM BP-927) and Streptomyces hygroscopicus subsp. yakushimaensis No. 7238 (FERM BP-928). Such macrolides are particularly numbered FR-900506, FR-900520, FR-900523 and FR-900525. And the preparation of some their derivatives is also described.

International Patent Application WO 89/05304, European Patent Application Nos. 353678, 349049, 349061, 356399, 402931, etc also disclose a number of macrocyclic immunosuppressive compounds.

We have now found a novel group of compounds which possess certain advantageous properties over those disclosed previously.

Thus, according to the invention, we provide a new compound of the following formula:



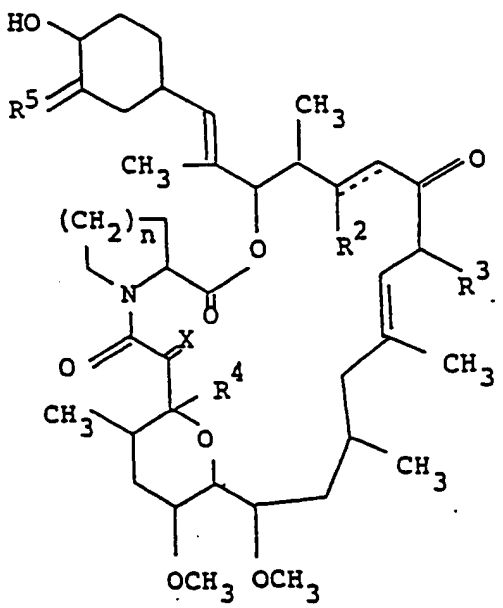
(I)

wherein R^1 is hydrogen, lower alkyl which may have one or more hydroxy, or aryl which may have suitable substituent(s),
R^2 is hydrogen, hydroxy or protected hydroxy,
R^3 is methyl, ethyl, propyl or allyl,
R^4 is hydroxy or alkoxy,
R^5 is oxo, (H, OH) or (H, alkoxy),
X is oxo or (H, OH),
n is an integer of 1 or 2, and
the symbol of a line and dotted line is a single bond or a double bond.

With respect to the tricyclo compounds (I) of this invention, it is to be understood that there may be one or more conformer(s) or stereoisomeric pairs such as optical and geometrical isomers due to asymmetric carbon atom(s) and double bond(s), and such isomers are also included within a scope of this invention.

According to this invention, the object tricyclo compounds (I) can be prepared by the following processes.

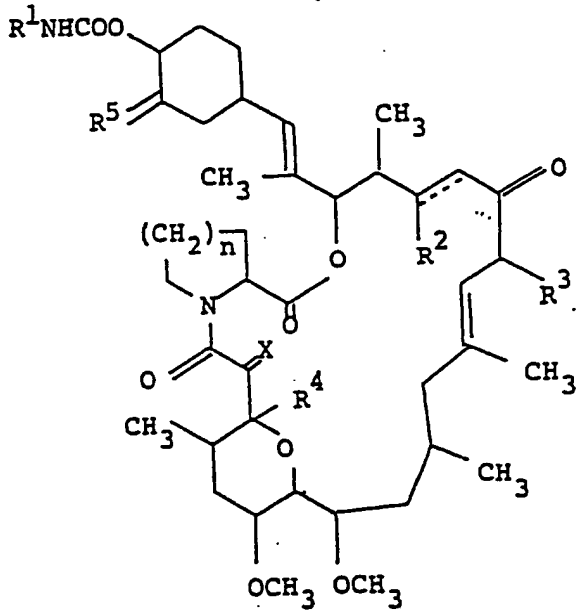
Process 1



(II)

or its reactive derivative
at the hydroxy group
or a salt thereof

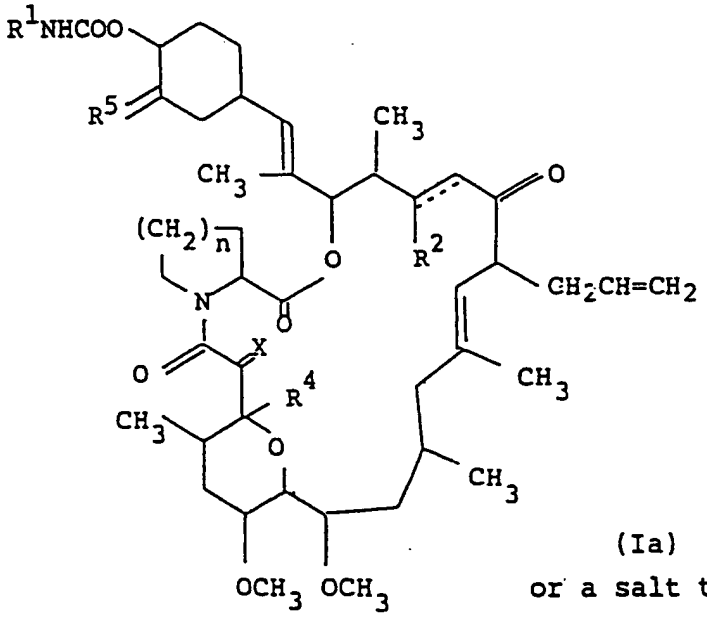
Introduction of R^1NHCO- group



(I)

or a salt thereof

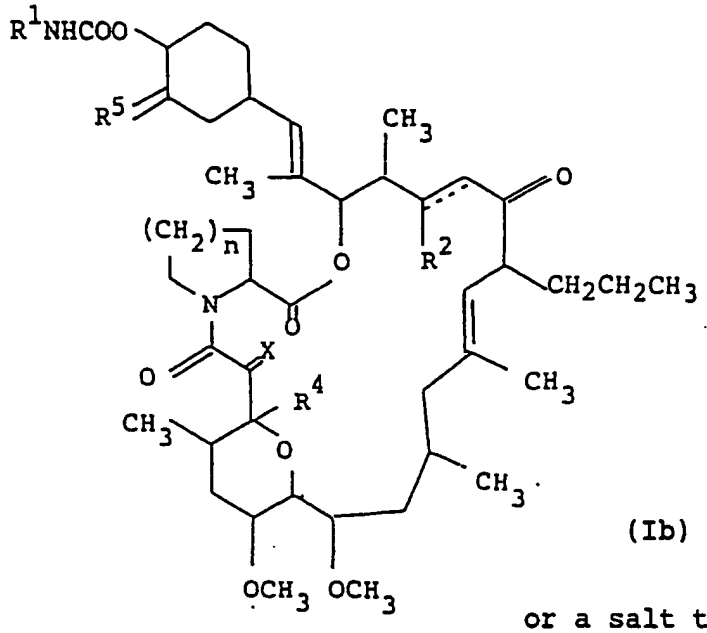
Process 2



(Ia)

or a salt thereof

Reduction



(Ib)

or a salt thereof

in which R^1, R^2, R^3, R^4, R^5, X and n are each as defined above.

Particulars of the above definitions and the preferred embodiments thereof are explained in detail as follows.

The term "lower" used in the specification is intended to mean 1 to 6 carbon atoms, unless otherwise indicated.

Suitable alkyl moiety in "alkoxy" and "lower alkyl which may have one or more hydroxy" may include a straight or branched lower alkyl such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, pentyl, hexyl, and the like. And particularly, the preferable "alkoxy" is methoxy, and the preferable "lower alkyl which may have one or more

hydroxy" is propyl or propyl having one or two hydroxy such as 2-hydroxypropyl and 2,3-dihydroxypropyl.

Suitable "aryl which may have suitable substituent(s)" may include phenyl, tolyl, xylyl, mesityl, naphthyl, and the like, which may have suitable substituent(s) such as halogen (e.g. fluoro, chloro, bromo, iodo), and the like.

Suitable hydroxy-protective group in the "protected hydroxy" may be the conventional ones including "R^6NHCO-" group, in which R^6 is the ones as defined in R^1.

The processes for production of tricyclo compounds (I) of this invention are explained in detail in the following.

Process 1 :

The compound (I) or a salt thereof can be prepared by introducing "R^1NHCO-" group into the compound (II) or its reactive derivative at the hydroxy group or a salt thereof.

Suitable introducing agent of "R^1NHCO-" group used in this reaction may be a conventional one such as carbamic acid and their reactive derivative, for example, an acid halide, an acid anhydride, an activated amide, an activated ester, isocyanate, and the like. Preferable example of such reactive derivative may include acid chloride, acid bromide, a mixed acid anhydride with an acid such as substituted phosphoric acid (e.g. dialkylphosphoric acid, phenylphosphoric acid, diphenylphosphoric acid, dibenzylphosphoric acid, halogenated phosphoric acid, etc.), dialkylphosphorous acid, sulfurous acid, thiosulfuric acid, sulfuric acid, alkyl carbonate (e.g. methyl carbonate, ethyl carbonate, propyl carbonate, etc.), aliphatic carboxylic acid (e.g.

pivalic acid, pentanoic acid, isopentanoic acid, 2-ethylbutyric acid, trichloroacetic trifluoroacetic acid, etc.), aromatic carboxylic acid (e.g. benzoic acid, etc.), a symmetrical acid anhydride, an activated acid amide with a heterocyclic compound containing imino function such as imidazole, 4-substituted imidazole, dimethylpyrazole, triazole and tetrazole, an activated ester (e.g. p-nitrophenyl ester, 2,4-dinitrophenyl ester, trichlorophenyl ester, pentachlorophenyl ester, mesylphenyl ester, phenylazophenyl ester, phenyl thioester, p-nitrophenyl thioester, p-cresyl thioester, carboxymethyl thioester, pyridyl ester, piperidinyl ester, 8-quinolyl thioester, or an ester with a N-hydroxy compound such as N,N-dimethylhydroxylamine, 1-hydroxy-2-(1H)-pyridone, N-hydroxysuccinimide, N-hydroxyphthalimide, 1-hydroxybenzotriazole, 1-hydroxy-6-chlorobenzotriazole, etc.), isocyanate, and the like.

Preferred embodiment of the introducing agent of "R^1NHCO-" thus defined may be represented by the following chemical formulae,

$$R^1\text{-}N{=}C{=}O \qquad \text{(IIIa)}$$

in which R^1 is as defined above, preferably aryl which may have suitable substituent(s), and

$$R^1\text{-}NH_2 \qquad \text{(IIIb)}$$

in which R^1 is as defined above, preferably hydrogen or lower alkyl which may have one or more hydroxy.

Suitable reactive derivative at the hydroxy group of the compound (II) may be conventional one which is capable of replacing a hydroxy group with "R^1NHCOO-" such as an activated ester as mentioned above.

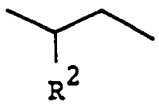
In this reaction, in case that the compound (II) is used in the form of an activated ester, it is preferable to use the compound (IIIb) as the introducing agent.

The reaction is preferably conducted in the presence of an organic or inorganic base such as alkali metal (e.g. lithium, sodium, potassium, etc.), alkaline earth metal (e.g. calcium, etc.), alkali metal hydride (e.g. sodium hydride, etc.), alkaline earth metal hydride (e.g. calcium hydride, etc.), alkali metal hydroxide (e.g. sodium hydroxide, potassium hydroxide, etc.), alkali metal carbonate (e.g. sodium carbonate, potassium carbonate, etc.), alkali metal hydrogen carbonate (e.g. sodium hydrogen carbonate, potassium hydrogen carbonate, etc.), alkali metal alkoxide (e.g. sodium methoxide, sodium ethoxide, potassium tert-butoxide, etc.), alkali metal alkanoic acid (e.g. sodium acetate, etc.), trialkylamine (e.g. triethylamine, etc.), pyridine compound (e.g. pyridine, lutidine, picoline, 4-N,N-dimethylaminopyridine, etc.), quinoline, and the like.

The reaction is usually conducted in a conventional solvent which does not adversely influence the reaction such as water, acetone, dichloromethane, alcohol (e.g. methanol, ethanol, etc.), tetrahydrofuran, pyridine, benzene, N,N-dimethylformamide, etc., or a mixture thereof, and further in case that the base or the introducing agent of the R^1NHCO- group is in liquid, it can also be used as a solvent.

The reaction temperature is not critical and the reaction is usually conducted under from cooling to heating.

This process includes, within a scope thereof, a case that during the reaction, the hydroxy group for R^2 of the compound (II) may occasionally be transformed into the corresponding "R^6NHCOO" group, in which R^6 is the ones as defined in R^1 in the object compound (I).

Further, this process also includes, within a scope thereof, a case that the compound (II) having a partial structure of the formula :  , wherein R^2 is hydroxy, may occasionally be eliminated during the reaction to give the compound (I) having a partial structure of the formula :  .

The starting compound (II) in the process 1 can be prepared in the same manner as described in the patent applications stated before, such as European Patent Application Nos. 184162 and 353678, International Patent Application WO 89/05304 and the like.

Process 2 :

The compound (Ib) or a salt thereof can be obtained by reducing the compound (Ia) or a salt thereof.

Reduction in this process can be conducted by a conventional method which is capable of reducing an allyl group to a propyl group, such as catalytic reduction, or the like.

Suitable catalysts used in catalytic reduction are conventional ones such as platinum catalysts (e.g. platinum plate, spongy platinum, platinum black, colloidal platinum, platinum oxide, platinum wire, etc.), palladium catalysts (e.g. spongy palladium, palladium black, palladium oxide, palladium on carbon, colloidal palladium, palladium on barium sulfate, palladium on barium carbonate, etc.), nickel catalysts (e.g. reduced nickel, nickel oxide, Raney nickel, etc.), cobalt catalysts (e.g. reduced cobalt, Raney cobalt, etc.), iron catalysts (e.g. reduced iron, Raney iron, etc.), copper catalysts (e.g. reduced copper, Raney copper, Ullman copper, etc.), and the like.

The reduction is usually conducted in a conventional solvent which does not adversely influence the reaction such as water, methanol, ethanol, propanol, pyridine, ethyl acetate, N,N-dimethylformamide, dichloromethane, or a mixture thereof.

The reaction temperature of this reduction is not critical and the reaction is usually conducted under from cooling to warming.

This process includes, within a scope thereof, a case that the double bond defined by the symbol of a line and dotted line may occasionally be reduced during the reaction to give a single bond.

PHARMACOLOGICAL ACTIVITIES OF THE TRICYCLO COMPOUNDS

The tricyclo compounds (I) possess pharmacological activities such as immunosuppressive activity, antimicrobial activity, and the like, and therefore are useful for the treatment and prevention of immune-mediated

diseases controlled by a immunosuppressant such as the resistance by transplantation of organs or tissue such as heart, kidney, liver, medulla ossium, skin, cornea, lung, pancreas, intestinum tenue, limb, muscle, nervus, etc.; graft-versus-host diseases by medulla ossium transplantation; autoimmune diseases such as rheumatoid arthritis, systemic lupus erythematosus, Hashimoto's thyroiditis, multiple sclerosis, myasthenia gravis, type I diabetes, and the like; and further infectious diseases caused by pathogenic microorganisms.

And further, the tricyclo compounds (I) are also useful in the topical administration for the treatment and the prophylaxis of inflammatory and hyperproliferative skin diseases and cutaneous manifestations of immunologically-mediated illnesses, such as, psoriasis, atopical dermatitis, contact dermatitis and further eczematous dermatitises, seborrhoeis dermatitis, Lichen planus, Pemphigus, bullous Pemphigoid, Epidermolysis bullosa, urticaria, angioedemas, vasculitides, erythemas, cutaneous eosinophilias, Lupus erythematosus and Alopecia areata.

Further, the compound (I) of the present invention is also useful for the treatment and prevention of; various diseases of the eye such as autoimmune diseases and so on (e.g. keratoconjunctivitis, vernal conjunctivitis, uveitis associated with Behcet's disease, keratitis, herpetic keratitis, conical cornea, dystrophia epithelialis corneae, leukoma, ocular pemphigus, Mooren's ulcer, Sclevitis, Graves' ophthalmopathy, etc.); reversible obstructive airways disease, which includes conditions such as asthma (e.g. bronchial asthma, allergic asthma, intrinsic asthma, extrinsic asthma and dust asthma), particularly chronic or inveterate asthma (e.g. late asthma and airway hyper-responsiveness), bronchitis and the like;

nephrotic syndrome such as glomerulonephritis;
hemolytic-uremic syndrome;
photoallergic sensitivity;
male pattern alopecia or alopecia senilis; etc.

And further, the compound (I) of the present invention is useful for various diseases because of its useful pharmaceutical activities such as liver regenerating activity, augmenting activity of chemotherapeutic effect, and so on.

As examples for showing such pharmacological activities, the pharmacological test data of the tricyclo compounds (I) is illustrated in the following.

Test 1

Suppression of in vitro Mixed Lymphocyte Reaction (MLR)

The MLR test was preformed in microtiter plates, with each well containing 5 x 10^5 C57BL/6 responder cells ($H\text{-}2^b$), 5 x 10^5 mitomycin C treated (25 µg/ml mitomycin C at 37°C for 30 minutes and washed three times with RPMI 1640 medium) BALB/C stimulator cells ($H\text{-}2^d$) in 0.2 ml RPMI 1640 medium supplemented with 10% fetal calf serum, 2 mM sodium bicarbonate, penicillin (50 unit/ml) and streptomycin (50 µg/ml). The cells were incubated at 37°C in humidified atmosphere of 5% carbon dioxide and 95% of air for 68 hours and pulsed with ^{3}H-thymidine (0.5 µCi) 4 hours before the cells were collected. The object compound of this invention were dissolved in ethanol and further diluted in RPMI 1640 medium and added to the cultures to give final concentrations of 100 nM or less.

The IC_{50} value (mol concentration to suppress 50% or MLR) was calculated by a conventional method, which is shown in the following Table 1.

Table 1 : IC_{50} value of MLR test on tricyclo compounds (I)

Test compound	IC_{50} value (mol/liter)
tricyclo compound prepared in Example 4-(2)	1.4×10^{-8}

The pharmaceutical composition of this invention can be used in the form of a pharmaceutical preparation, for example, in solid, semisolid or liquid form, which contains the tricyclo compounds (I), as an active ingredient, in admixture with an organic or inorganic carrier or excipient suitable for external, enteral or parenteral applications. The active ingredient may be compounded, for example, with the usual non-toxic, pharmaceutically acceptable carriers for tablets, pellets, capsules, suppositories, solutions, emulsions, suspensions, injections, ointments, liniments, eye drops lotion, gel, creme and any other form suitable for use. The carriers which can be used are water, glucose, lactose, gum acacia, gelatin, mannitol, starch paste, magnesium trisilicate, talc, corn starch, keratin, colloidal silica, potato starch, urea and other carriers suitable for use in manufacturing preparations, in solid, semisolid, or liquid form, and in addition auxiliary, stabilizing, thickening, solubilizing an coloring agents and perfumes may be used. Particularly, as a solubilizing agent, there may be exemplified water-soluble cellulose polymer (i.e. hydroxypropyl methylcellulose, etc.), water-soluble glycol (i.e. propylene glycol, etc.), etc. The active object compound is included in the pharmaceutical composition in an amount sufficient to produce the desired effect upon the process or condition of diseases.

For applying this composition to human, it is preferable to apply it by parenteral or enteral administration. While the dosage of therapeutically effective amount of the tricyclo compound (I) varies from and also depends upon the age and condition of each individual patient to be treated, a daily dose of about 0.01-1000 mg, preferably 0.1-500 mg and more preferably 0.5-100 mg, of the active ingredient is generally given for treating diseases, and an average single dose of about 0.5 mg, 1 mg, 5 mg, 10 mg, 50 mg, 100 mg, 250 mg and 500 mg is generally administered.

The following examples are given for the purpose of illustrating the present invention.

Example 1

To a mixture of 17-allyl-1-hydroxy-12-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo-[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (1.0 g) and pyridine (1.77 g) in anhydrous dichloromethane (10 ml) was added phenyl isocyanate (1.28 g), and the mixture was stirred for 16 hours at ambient temperature. The reaction mixture was washed with 1N aqueous hydrochloric acid solution, water, aqueous sodium bicarbonate solution and brine successively, and dried over magnesium sulfate. The solvent was evaporated in vacuo and the residue was purified by silica gel column chromatography, which was eluted with a mixture of dichloromethane and diethyl ether (2:1 V/V) to give 17-allyl-1-hydroxy-23,25-dimethoxy-12-[2-(3-methoxy-4-phenylcarbamoyloxycyclohexyl)-1-methylvinyl]-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (1.01 g).

FAB-MS : m/z 927 (M^+ + Na)

Example 2

To a mixture of 17-allyl-1,14-dihydroxy-12-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (400 mg) and pyridine (400 mg) in anhydrous dichloromethane (4 ml) was added p-nitrophenyl chloroformate (300 mg), and the mixture was stirred for one hour at ambient temperature. To the mixture was added 3-amino-1-propanol (300 mg) and after stirred for one hour at ambient temperature, additional portion of 3-amino-1-propanol (150 mg) was added. After stirred for 30 minutes, the mixture was washed with brine, dried over magnesium sulfate, and evaporated in vacuo. The residue was purified by silica gel column chromatography, which was eluted with ethyl acetate to give 17-allyl-1-hydroxy-12-[2-[4-(3-hydroxypropylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo-[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (96 mg).

FAB-MS : 909 (M^+ + Na)

mp : 92-93°C

Further elution of the column with a mixture of ethyl acetate and methanol (98-2 V/V %) afforded 17-allyl-1-hydroxy-14-(3-hydroxypropylcarbamoyloxy)-12-[2-[4-(3-hydroxypropylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (133 mg).

FAB-MS : 1028 (M^+ + Na)

Example 3

A solution of 17-allyl-1-hydroxy-12-[2-[4-(3-hydroxypropylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (50 mg) in acetic acid (2 ml) was suspended with 5% palladium on carbon (25 mg), and the reaction mixture was stirred for 4 hours under hydrogen atmosphere at one atm. The catalyst was filtered off and the filtrate was concentrated in vacuo. The residue was purified by flash chromatography with silica gel, which was eluted with ethyl acetate to give 1-hydroxy-12-[2-[4-(3-hydroxypropylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (35 mg) as white powders.

FAB-MS : 913 (M^+ + Na)

mp : 81-83°C

Example 4

The following compounds were obtained by reacting the corresponding starting compounds with isocyanate compounds according to a similar manner to that of Example 1.

(1) 17-Allyl-12-[2-[4-(4-fluorophenylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-1-hydroxy-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (Yield : 91%).

FAB-MS : m/z 946 (M^+ + Na)

(2) 17-Allyl-12-[2-[4-(4-chlorophenylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-1,14-dihydroxy-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (Yield : 15%).

FAB-MS : 979 (M^+ + Na)

(3) 17-Allyl-12-[2-[4-(4-chlorophenylcarbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-1-hydroxy-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (Yield : 75%).

FAB-MS : 963 (M^+ + Na)

Example 5

The following compounds were obtained by subjecting 17-allyl-1,14-dihydroxy-12-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone to a similar reaction to that of Example 2.

(1) 17-Allyl-1-hydroxy-23,25-dimethoxy-12-[2-(3-methoxy-4-propylcarbamoyloxycyclohexyl)-1-methylvinyl]-13,19,21-27-tetramethyl-11,28-dioxa-4-azatricyclo-[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (Yield : 21%).

FAB-MS : m/z 893 (M^+ + Na)

(2) 17-Allyl-1-hydroxy-12-[2-[4-[(2S)-2-hydroxypropylcarbamoyloxy]-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (Yield : 12%).

FAB-MS : m/z 909 (M^+ + Na)

(3) 17-Allyl-1-hydroxy-12-[2-[4-(2,3-dihydroxypropyl-carbamoyloxy)-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-aza-tricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (Yield : 2%).

FAB-MS : m/z 925 (M^+ + Na)

(4) 17-Allyl-1-hydroxy-12-[2-[4-[(2R)-2-hydroxypropyl-carbamoyloxy]-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-14,18-diene-2,3,10,16-tetraone (Yield : 43%).

FAB-MS : 909 (M^+ + Na)

mp : 94-96°C

Example 6

12-[2-(4-Carbamoyloxy-3-methoxycyclohexyl)-1-methylvinyl]-1-hydroxy-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (49 mg) was obtained by subjecting 1-hydroxy-12-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (130 mg) to a similar reaction to that of Example 2.

FAB-MS : m/z 856 (M^+ + Na)

Example 7

The following compounds were obtained according to a similar manner to that of Example 3.

(1) 1-Hydroxy-12-[2-[4-[2(2R)-2-hydroxypropyl-carbamoyloxy]-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-

azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (Yield : 40%).

FAB-MS : 913 (M^+ + Na)

(2) 1-Hydroxy-23,25-dimethoxy-12-[2-(3-methoxy-4-propylcarbamoyloxycyclohexyl)-1-methylvinyl]-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-azatricyclo-[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (Yield : 39%).

FAB-MS : m/z 897 (M^+ + Na)

(3) 1-Hydroxy-12-[2-[4-[(2S)-2-hydroxypropylcarbamoyloxy]-3-methoxycyclohexyl]-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-propyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (Yield : 84%).

FAB-MS : m/z 913 (M^+ + Na)

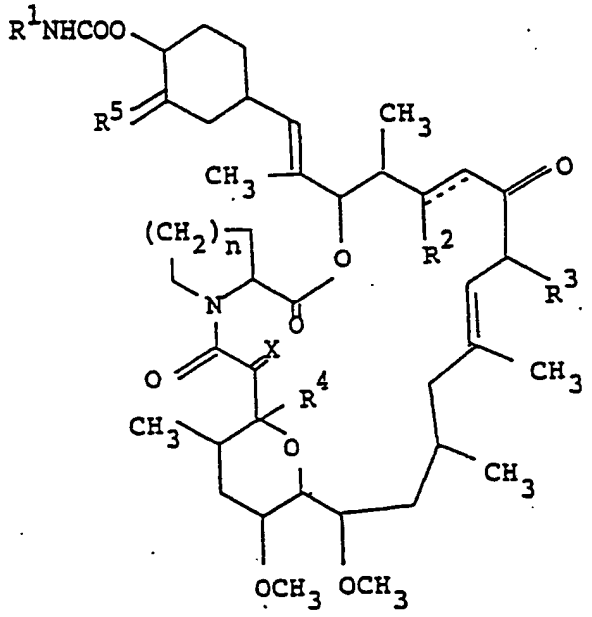
Example 8

12-[2-(4-Carbamoyloxy-3-methoxycyclohexyl)-1-methylvinyl]-1-hydroxy-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-allyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (33 mg) was obtained by subjecting 1-hydroxy-12-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylvinyl]-23,25-dimethoxy-13,19,21,27-tetramethyl-11,28-dioxa-17-allyl-4-azatricyclo[22.3.1.$0^{4,9}$]octacos-18-ene-2,3,10,16-tetraone (80 mg) to a similar reaction to that of Example 2.

FAB-MS : m/z 853 (M^+ + Na)

CLAIMS :

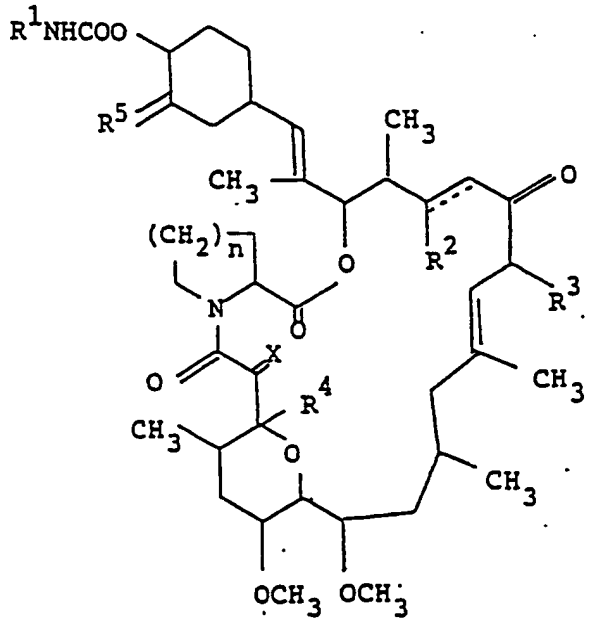
1. A compound of the formula :



wherein R^1 is hydrogen, lower alkyl which may have one or more hydroxy, or aryl which may have suitable substituent(s),
R^2 is hydrogen, hydroxy or protected hydroxy,
R^3 is methyl, ethyl, propyl or allyl,
R^4 is hydroxy or alkoxy,
R^5 is oxo, (H, OH) or (H, alkoxy),
X is oxo or (H, OH),
n is an integer of 1 or 2, and
the symbol of a line and dotted line is a single bond or a double bond,
and salts thereof.

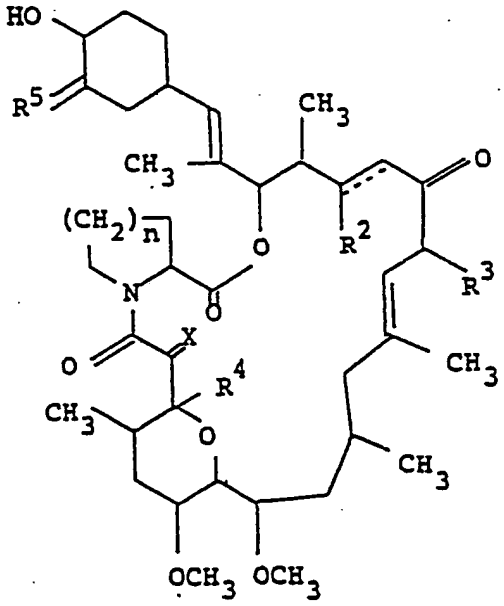
2. The compound of Claim 1, wherein R^2 and R^4 are hydroxy, R^3 is allyl, R^5 is (H, methoxy), X is oxo, n is 2 and the symbol of a line and dotted line is a single bond.

3. A Process for preparing a compound of the formula :



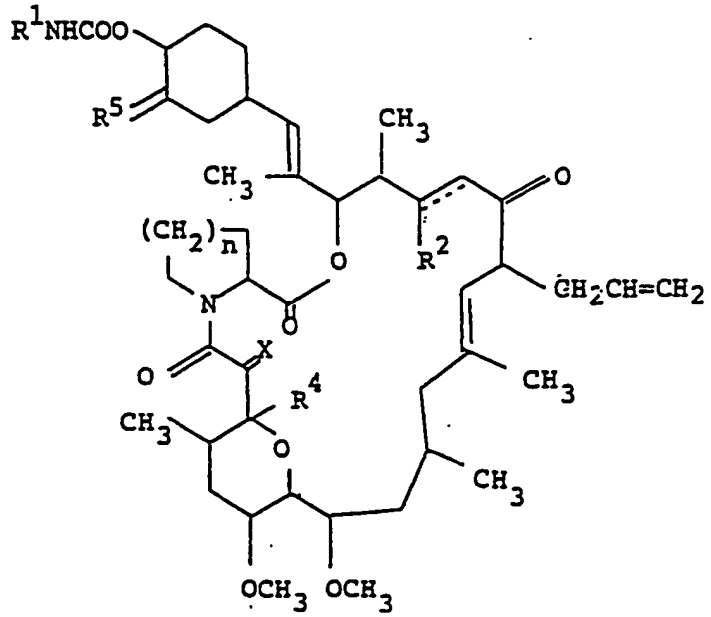
wherein R^1 is hydrogen, lower alkyl which may have one or more hydroxy, or aryl which may have suitable substituent(s),
R^2 is hydrogen, hydroxy or protected hydroxy,
R^3 is methyl, ethyl, propyl or allyl,
R^4 is hydroxy or alkoxy,
R^5 is oxo, (H, OH) or (H, alkoxy),
X is oxo or (H, OH),
n is an integer of 1 or 2, and
the symbol of a line and dotted line is a single bond or a double bond,
or a salt thereof,
which comprises

(a) subjecting a compound of the formula :

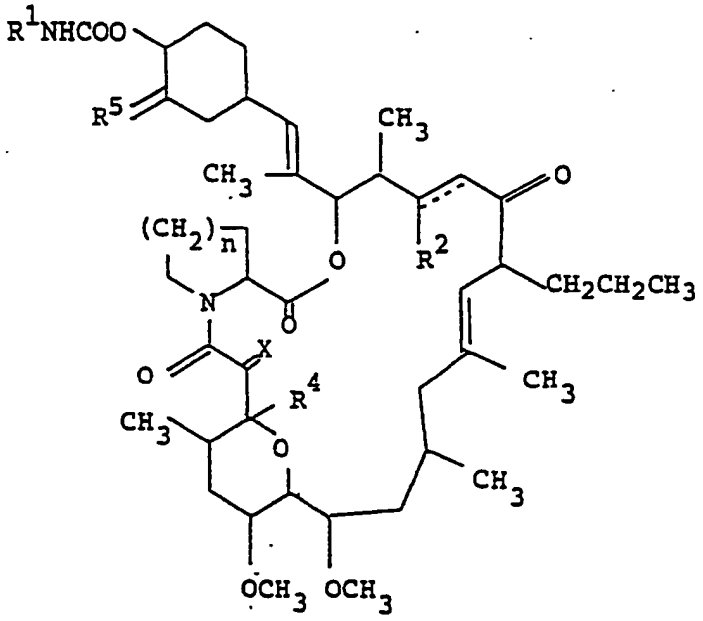


wherein R^2, R^3, R^4, R^5, X, n and the symbol of a line and dotted line are each as defined above, or its reactive derivative at the hydroxy group or a salt thereof, to introduction reaction of R^1NHCO- group, or

(b) reducing a compound of the formula :



wherein R^1, R^2, R^4, R^5, X, n and the symbol of a line and dotted line are each as defined above,
or a salt thereof, to give a compound of the formula:



wherein R^1, R^2, R^4, R^5, X, n and the symbol of a line and dotted line are each as defined above,
or a salt thereof.

4. A pharmaceutical composition which comprises a compound of Claim 1 and a pharmaceutically acceptable carrier or excipient.

5. A process for preparing a pharmaceutical composition which comprises admixing a compound of Claim 1 with a pharmaceutically acceptable carrier or excipient.

6. A use of a compound of Claim 1 as a medicament.

7. A use of a compound of Claim 1 as an immunosuppressant or an antimicrobial agent.

8. A use of a compound of Claim 1 for manufacturing a medicament for treating immune-mediated diseases or infectious diseases.

9. A method for treating or preventing immune-mediated diseases or infectious diseases which comprises administering a compound of claim 1 to human or animal.

10. A medicament for treating immune-mediated diseases or infectious diseases which comprises, as an active ingredient, a compound of Claim 1.

FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

V. [X] OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE [1]

This international search report has not been established in respect of certain claims under Article 17(2) (a) for the following reasons:

1. [X] Claim numbers 9, because they relate to subject matter not required to be searched by this Authority, namely:

See PCT Rule 39.1(iv):
methods for treatment of the human or animal body by surgery or therapy, as well as diagnostic methods.

2. [ ] Claim numbers ....., because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claim numbers ....., because they are dependent claims and are not drafted in accordance with the second and third sentences of PCT Rule 6.4(a).

VI. [ ] OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING [2]

This International Searching Authority found multiple inventions in this international application as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims of the international application.

2. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims of the international application for which fees were paid, specifically claims:

3. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:

4. [ ] As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

Remark on Protest

[ ] The additional search fees were accompanied by applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

V. [X] OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND UNSEARCHABLE [1]

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3. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:

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[ ] The additional search fees were accompanied by applicant's protest.

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ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO. JP 9100314
SA 45191

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report.
The members are as contained in the European Patent Office EDP file on 26/06/91
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